REPORT DOCUMENTATION PAGE

Form Approved OMB NO. 0704-0188

1. REPORT DATE (DD-MM-YYYY)	2. REPORT TYPE	3. DATES COVERED (From - To)
14-09-2012	Final Report	1-Apr-2011 - 31-Mar-2012

4. TITLE AND SUBTITLE

Final Report
Bioelectrochemical Systems Workshop:
Standardized Analyses, Design Benchmarks, and Reporting

5a. CONTRACT NUMBER
W911NF-11-1-0148

5b. GRANT NUMBER

5c. PROGRAM ELEMENT NUMBER
622720

6. AUTHORS
John M. Regan

5d. PROJECT NUMBER

5e. TASK NUMBER

5f. WORK UNIT NUMBER

7. PERFORMING ORGANIZATION NAMES AND ADDRESSES

Pennsylvania State University
Office of Sponsored Programs
The Pennsylvania State University
University Park, PA 16802 -7000

8. PERFORMING ORGANIZATION REPORT NUMBER

9. SPONSORING/MONITORING AGENCY NAME(S) AND ADDRESS(ES)

U.S. Army Research Office
P.O. Box 12211
Research Triangle Park, NC 27709-2211

10. SPONSOR/MONITOR'S ACRONYM(S)
ARO

11. SPONSOR/MONITOR'S REPORT NUMBER(S)
59850-EV.1

12. DISTRIBUTION AVAILIBILITY STATEMENT

Approved for Public Release; Distribution Unlimited

13. SUPPLEMENTARY NOTES

The views, opinions and/or findings contained in this report are those of the author(s) and should not contrued as an official Department of the Army position, policy or decision, unless so designated by other documentation.

14. ABSTRACT

Bioelectrochemical systems (BESs) have attracted a considerable amount of interest in the past decade in part because of their potential to transform waste treatment into an energy-neutral or even producing endeavor. Research on these systems has greatly advanced our understanding of microbially catalyzed extracellular electron transfer, and design improvements have significantly improved power densities in a short period of time. These dramatic improvements in system performance and the current initiatives in scaling up these systems have all

15. SUBJECT TERMS

microbial fuel cells, standard testing, electrochemistry

16. SECURITY CLASSIFICATION OF:			17. LIMITATION OF ABSTRACT	15. NUMBER OF PAGES	19a. NAME OF RESPONSIBLE PERSON
a. REPORT	b. ABSTRACT	c. THIS PAGE			John Regan
UU	UU	UU	UU		19b. TELEPHONE NUMBER: 814-865-9436

Report Title

Final Report
Bioelectrochemical Systems Workshop:
Standardized Analyses, Design Benchmarks, and Reporting

ABSTRACT

Bioelectrochemical systems (BESs) have attracted a considerable amount of interest in the past decade in part because of their potential to transform waste treatment into an energy-neutral or even producing endeavor. Research on these systems has greatly advanced our understanding of microbially catalyzed extracellular electron transfer, and design improvements have significantly improved power densities in a short period of time. These dramatic improvements in system performance and the current initiatives in scaling up these systems have all occurred in the absence of standard design and testing procedures, and this lack of standardization hinders the pace of technology development. On September 14 and 15, 2011, an Army-sponsored workshop was convened at The Pennsylvania State University in University Park, Pennsylvania with the following goals: (1) identify BES design and performance parameters that can be meaningfully standardized, (2) determine the best practices for reliable comparative testing of these parameters, and (3) develop a benchmarking framework from which subsequent advancements can be uniformly compared. The workshop had 30 participants, including many of the leading BES researchers in academia from the United States, Canada, Australia, the Netherlands, and Belgium; individuals from the Department of Defense; and representatives from five companies with existing BES initiatives.

Enter List of papers submitted or published that acknowledge ARO support from the start of the project to the date of this printing. List the papers, including journal references, in the following categories:

(a) Papers published in peer-reviewed journals (N/A for none)

Received Paper

TOTAL:

Number of Papers published in peer-reviewed journals:

(b) Papers published in non-peer-reviewed journals (N/A for none)

Received Paper

TOTAL:

Number of Papers published in non peer-reviewed journals:

(c) Presentations

Number of Presentations: 0.00

Non Peer-Reviewed Conference Proceeding publications (other than abstracts):

Received Paper

TOTAL:

Number of Non Peer-Reviewed Conference Proceeding publications (other than abstracts):

Peer-Reviewed Conference Proceeding publications (other than abstracts):

Received Paper

TOTAL:

Number of Peer-Reviewed Conference Proceeding publications (other than abstracts):

(d) Manuscripts

Received Paper

TOTAL:

Number of Manuscripts:

Books

Received Paper

TOTAL:

Patents Submitted

Patents Awarded

Awards

Graduate Students

NAME	PERCENT_SUPPORTED
FTE Equivalent:	
Total Number:	

Names of Post Doctorates

NAME	PERCENT_SUPPORTED
FTE Equivalent:	
Total Number:	

Names of Faculty Supported

NAME	PERCENT_SUPPORTED
FTE Equivalent:	
Total Number:	

Names of Under Graduate students supported

NAME	PERCENT_SUPPORTED
FTE Equivalent:	
Total Number:	

Student Metrics

This section only applies to graduating undergraduates supported by this agreement in this reporting period

The number of undergraduates funded by this agreement who graduated during this period:	0.00
The number of undergraduates funded by this agreement who graduated during this period with a degree in science, mathematics, engineering, or technology fields:.....	0.00
The number of undergraduates funded by your agreement who graduated during this period and will continue to pursue a graduate or Ph.D. degree in science, mathematics, engineering, or technology fields:.....	0.00
Number of graduating undergraduates who achieved a 3.5 GPA to 4.0 (4.0 max scale):	0.00
Number of graduating undergraduates funded by a DoD funded Center of Excellence grant for Education, Research and Engineering:	0.00
The number of undergraduates funded by your agreement who graduated during this period and intend to work for the Department of Defense	0.00
The number of undergraduates funded by your agreement who graduated during this period and will receive scholarships or fellowships for further studies in science, mathematics, engineering or technology fields:	0.00

Names of Personnel receiving masters degrees

NAME

Total Number:

Names of personnel receiving PHDs

NAME

Total Number:

Names of other research staff

NAME PERCENT_SUPPORTED

FTE Equivalent:

Total Number:

Sub Contractors (DD882)

Inventions (DD882)

Scientific Progress

Introduction

Bioelectrochemical systems (BESs) have attracted a considerable amount of interest in the past decade in part because of their potential to transform waste treatment into an energy-neutral or even producing endeavor. This technology would therefore enable remote waste treatment without the need for energy provision, making it especially suitable for applications such as wastewater treatment at forward base camps. Research on these systems has greatly advanced our understanding of microbially catalyzed extracellular electron transfer, and design improvements have improved power densities by approximately six orders of magnitude [1], from an early design that used a salt bridge for electrochemical continuity [2] to newer designs that greatly reduce ohmic constraints and increase specific surface areas of the electrodes. The development of this technology is just beginning to transition to pilot-scale testing, with three different designs field tested for treating brewery, winery, and municipal wastewaters [3].

These dramatic improvements in system performance and the current initiatives in scaling up these systems have all occurred in the absence of standard design and testing procedures. Each research group has its preferred designs, media, inoculation and enrichment strategies, and performance evaluations. As a result, it is very difficult to properly compare the results of different studies, as each research team uses entirely different conditions and analyses. This lack of standardization hinders the pace of technology development. It is anticipated that the establishment of benchmarking conditions for BESs will accelerate the commercialization of these systems, just as experienced by other technologies such as hydrogen fuel cells with the development of the U.S. Fuel Cell Council (now the Fuel Cell and Hydrogen Energy Association).

On September 14 and 15, 2011, an Army-sponsored workshop was convened at The Pennsylvania State University in University Park, Pennsylvania with the following goals: (1) identify BES design and performance parameters that can be meaningfully standardized, (2) determine the best practices for reliable comparative testing of these parameters, and (3) develop a benchmarking framework from which subsequent advancements can be uniformly compared. The workshop had 30 participants, including many of the leading BES researchers in academia from the United States, Canada, Australia, the Netherlands, and Belgium; individuals from the Department of Defense; and representatives from five companies with existing BES initiatives. The workshop was opened with a presentation from Jerry Miller, who provided an overview of ERDC and ERDC investments that could benefit from standardization of BES technology. This was followed with invited presentations that addressed microbial and biofilm characterizations and considerations, materials considerations, chemical and electrochemical processes and monitoring, performance metrics and control, alternative applications, and system design and operation. Following the technical content of the presentations, the workshop transitioned to group discussions on benchmark designs, monitoring and characterization techniques, and reporting requirements. Appendix 1 shows the workshop agenda.

Microbial Considerations

BES research can be segregated into pure- and mixed-culture studies. Pure cultures are frequently used to investigate fundamental processes involved in extracellular electron transfer, while mixed cultures enable the testing of system performance in nonsterile environmental settings that involve cooperation and competition of exoelectrogenic and nonexoelectrogenic microbial populations (Figure 1, see attachment), such as those encountered in wastewater- or sediment-fed BESs. The discussion of microbial considerations begins with this segregation, as the relevant questions and techniques are frequently distinct between these two types of studies.

In mixed-culture BES studies, the main microbial focus is typically related to characterizing the anode biofilm community composition, and less frequently also including the suspended community. The intent of these studies is to identify the predominant community members, with the presumption that this is related to the exoelectrogenic biofilm activity, and to investigate whether the community structure is a function of design and operational parameters such as inoculum, substrate [4], anode material [5], anode potential [6, 7], and operational mode [8]. There are numerous tools available for characterizing microbial communities. The questions that were addressed include which tools are most suitable and informative for BES research, and when and where should biofilm samples be collected?

The most prevalent methods of community characterization in BES studies have entailed phylogenetic characterization based on 16S rRNA gene sequences. It is acknowledged that the interpretation of rRNA-inferred communities can be distorted by the absence of function associated with this genetic marker, and in methods that involve polymerase chain reaction (PCR) amplification the quantitative representation of community composition can be biased by factors such as differential DNA extraction, different rRNA operon copy numbers [9], and primer bias. Nevertheless, given the diversity of mechanisms involved in anode reduction, the lack of a universal molecular target to capture these mechanisms, the presence and contribution of nonexoelectrogenic populations, and the likely presence of unknown and uncharacterized exoelectrogens in these communities, the properly qualified use of phylogeny-targeted techniques offers a tractable approach to unravel this complexity and has enabled significant progress in our understanding of these complex communities. (This issue is common among all mixed-culture microbial ecology studies and is not at all unique to BESs.)

The significant majority of BES microbial community research has used either denaturing gradient gel electrophoresis (DGGE) or clone libraries to analyze amplified 16S rRNA genes. Both of these techniques have strengths and weaknesses that make them suitable for different types of characterization studies. As post-PCR methods, they both share the potential introduction of biased population recovery for the reasons mentioned above. DGGE is most appropriately used for the screening of community differences over time [10, 11] or from different treatments [7, 12]. The subjective comparison of DGGE fingerprints can be addressed through methods such as principal component analysis, which can be automated and also informative about the conditions that correlate with observed community differences [6]. Problems associated with this

technique include the comigration of distinct bands, thereby masking diversity; and the need for band excision to ascertain identity, which involves the subjective process of band selection followed by the additional effort of cloning and sequencing. If the primary interest is in identifying only those populations that change with different treatments, DGGE might be appropriate. However, if broad community characterization is the goal [4, 8], the analysis of clone libraries makes more sense because the use of DGGE for this purpose requires clone libraries anyway and constrains the sequences that can be recovered to those that are in the visible and distinct DGGE bands. The main concern with clone libraries is the number of cloned sequences necessary to adequately characterize the community. Rarefaction curves [13] and statistical metrics [14] are standard tools to estimate the extent of community coverage, but there seems to be no general agreement in molecular microbial ecology on what constitutes sufficient community characterization.

Pyrosequencing is a technique that addresses the issue of clone library coverage. Depending on the degree of sample multiplexing, this technique generates thousands to tens of thousands of sequences per sample. Pyrosequencing has not been used extensively in BES studies, but it is beginning to make inroads [15-17]. The initial introduction of this sequencing technology only permitted very short reads that did not provide sufficient resolution in phylogenetic identification. This has been overcome with technology advancements, which now enable several hundred basepairs of quality sequence per read. Cost has also been a limiting factor, but it is comparable in cost to an extensive cloning and sequencing effort and is certainly less expensive on a per sequence basis than cloning. Pyrosequencing offers a much deeper level of community coverage than cloning, allowing the recovery of sequences from rare populations in the community that would likely go undetected in a clone library or DGGE analysis. However, it is unknown whether those rare populations play a significant role in the community ecology, and it is interesting to note that pyrosequencing-derived rarefaction curves generally do not show improved asymptotic coverage of the community diversity.

Fluorescent in-situ hybridization (FISH) is a complementary technique that allows examination of specific subsets of a community, but is not suitable for high-resolution community characterization. FISH has been used sparingly in BES studies [8, 18-20]. It is particularly useful for examining issues related to localization within an aggregate or biofilm, which might be relevant in BESs to investigate whether exoelectrogenic populations are stratified with respect to distance from the anode or uniformly distributed across the biofilm thickness. Moreover, FISH does not introduce the quantitative bias associated with PCR-based techniques, making it a good technique for confirming end point PCR-derived quantitation.

Two other issues related to BES biofilm community sampling are when and where to sample. If the primary interest is in characterizing the stable community composition and not the acclimation dynamics, several studies reported anode biofilm stability by approximately month two of closed-circuit system operation [10, 21, 22]. Regarding where to sample, there has been very little reported on the potential spatial heterogeneity of biofilm communities in these systems, and most studies do not even provide specifics on the sampling location. A recent study [23] looked into community heterogeneity in BES anodes, using pyrosequencing to compare the communities in different locations on a planar carbon-cloth anode and throughout a volumetric graphite-brush anode, in which one would expect to find gradients [24] of pH and perhaps substrate and dissolved oxygen concentration. The data showed virtually identical communities in all locations of both systems, suggesting that sampling location may not be critical, at least in smaller lab-scale reactors.

In addition to community composition, it is often necessary to measure total biomass concentration in the biofilm. It has been demonstrated that biofilm architecture develops over the first several weeks of operation, with a corresponding improvement in system performance [11]; that the biofilm architecture is a function of external resistance and anode potential [7]; and that there is a direct correlation between biomass concentration and current production in Geobacter sulfurreducens biofilms [25]. Therefore, total biomass is necessary to properly characterize this transient acclimation period.

Biomass-normalized specific rates of substrate utilization and current generation are also essential for a fair comparison of different communities or pure cultures, where unmeasured differences in biomass density might give different apparent rates. Several biomass measurement techniques have been used in BES research, including total protein, dry weight, quantitative real-time PCR [6, 26], and total and viable cell counts following nucleic acid staining [11]. It is important to remember when using microscopy-based techniques that porous anode materials can harbor cells within the anode matrix that will not be visualized.

Pure-culture studies allow the investigation of exoelectrogenic and exoelectrotrophic activities without the complications associated with the biofilm harboring microbial populations engaged in other metabolisms or with distinct physiological and kinetic attributes. This opens up opportunities for comparing the relative performances of different exoelectrogenic cultures, with the total biomass measurements such as protein, biovolume, and thickness only including biomass that is relevant to the electrode reaction. Pure cultures also enable the isolation of the role of a specific gene product through the differential activities of wild type and mutant strains, and looking at transcriptomic and proteomic responses to different conditions. These types of studies with Geobacter have shown a significant upregulation in the pilin gene pilA and the outer membrane cytochrome genes omcB and omcZ in current-generating biofilms, an order of magnitude lower current production in pilA- and omcZ- strains [27], and changes in the spatial and temporal expression of omcZ during biofilm development. There are also a number of potential exoelectrotrophic BES applications, such as bioreduction of nitrate, U(VI), and chlorinated compounds as well as microbial electrosynthesis to convert carbon dioxide into organic products [28]. These reactions have been conclusively demonstrated in pure-culture systems, with reductive dechlorination being demonstrated with Anaeromyxobacter dehalogenans [29] and Geobacter lovleyi [30] and a comparative assessment of the exoelectrotrophic productivities of various acetogenic species [31].

There are two main strategies that have been used to examine heterogeneity in pure-culture exoelectrogenic biofilms, destructive sampling followed by microtoming to physically section the biofilm layers [32] and in situ examination using a

mini-MFC system coupled with confocal microscopy [33]. In the former approach, which included a microarray analysis of differential rRNA expression in the inner and outer layers of the biofilm, it was noted that the presentation of microarray data is typically insufficient in review, and the functional inferences should be demonstrated throughout knockout and complementation experiments. The in situ approach has been used to experimentally demonstrate acidification of the biofilm interior [33] and spatial and temporal expression of omcZ and citrate synthase using a short half-life GFP reporter system. Another modified BES design that incorporates a partitioned gold anode with a non-conductive gap has been used to measure biofilm conductivity [34], with results showing the conductance of current-producing Geobacter biofilms being similar to conductive polymers. Biofilm conductivity experiments should include temperature dependence to indicate the likely mechanism, as tunneling, hopping, and metallic mechanisms all have different temperature responses.

Certain biofilm characterization techniques are amenable to either mixed- or pure-culture applications, though their interpretation is always simplified in the latter case. Microelectrodes are extremely useful in measuring condition profiles within and adjacent to biofilms in lab-scale systems, and they have been used by several BES researcher groups. Oxygen crossover in air-cathode layered electrode assemblies was measured using oxygen microelectrodes to show that oxygen levels at the anode were consistently below 0.3 mg/L [35]. Redox and pH profiles were measured in Shewanella oneidensis MR-1 biofilms to show that anode reduction by this bacterium is not redox controlled [36]. Microelectrodes have also been developed and demonstrated with S. oneidensis MR-1 biofilms for the quantification of the electron-transfer mediating flavins [37], though this demonstration was not in a BES. Similarly, nuclear magnetic resonance has also been used with S. oneidensis MR-1 biofilms to study heterogeneity in effective diffusivity [38], but this demonstration was not performed in a BES system. These tools allow the inspection of chemical and physical gradients within biofilm systems, and not just the bulk conditions, to characterize microenvironments that influence and define microbial communities and activities in BES biofilms.

In studying microbial kinetics in BESs, there are a number of important system factors that can have a marked impact on the results. Current productivity is typically expressed as an electrode surface area-specific current density. For comparative studies on electrode materials, it may be necessary to use a projected surface area to calculate current density, at least in the absence of an accepted method of measuring microbially accessible electrochemically active surface area. It has been shown even with variably polished glassy carbon electrodes that the surface roughness of the electrode significantly influences the current density [39]. Therefore, it is recommended that fundamental studies of biofilm kinetics use polished (> P1500) planar electrodes and not porous electrodes as is often reported. Medium composition also affects biofilm kinetics, which is further complicated by changes in concentration for some components during microbial growth. This has been repeatedly demonstrated for pH [40], yet it is fairly common for the effluent pH to not be measured and reported. It is critical in BES studies to report the medium composition and concentration, including the solution conductivity, as well as the effluent bulk pH. Finally, the proper collection of anode kinetic data requires that the cathode not influence the rates. For example, oxygen intrusion in an air-cathode system induces a current associated with the production of hydrogen peroxide. This can be overcome by running the system as a three-electrode microbial electrolysis cell (MEC), with a two-chamber system to eliminate hydrogen consumption and enable an electron balance.

Catalyst Binder and Polymer Research

Some of the principle objectives of material research in BESs include developing durable polymer materials that resist biofouling and have low charge transfer and solution resistances. These materials must also have the potential to be produced inexpensively in large quantities. Presently Nafion® stands as the benchmark among metal catalyst binders and should be used as a reference system for new materials. Sulfonated cathode catalyst binders show increasing charge transfer resistance and lower power production with increased degree of sulfonation [41]. Neutral hydrophilic binders show lower initial power densities than Nafion® but they are stable and have comparable performance to Nafion® after over a month of operation [42]. With these results, and complemented by electrochemical impedance spectroscopy (EIS) analyses, a more rational design basis for binders is beginning to emerge that balances oxygen diffusion to the cathode and fouling and wetting of the cathode pore matrix.

Electrochemical Monitoring

A recurrent theme during the workshop was the importance of properly using and maintaining reference electrodes. One common problem, particularly with Ag/AgCl electrodes, is that they drift over time. This can introduce considerable error for long-term experiments. Drift should be corrected by using a reliable paired reference electrode in saturated KCl. The correct conversion to SHE should also be reported for the electrolyte used in the experiments, with the caution that 0.197 V vs SHE is only true for saturated KCl. This conversion can be determined for your experimental medium by comparing two Ag/AgCl electrodes in a two-chamber cell separated by a membrane, with one chamber filled with the medium and the other filled with a KCl solution having a reported conversion to SHE. Another consideration is accounting for the IR drop between the reference electrode and the working electrode, which increases with increasing current density and separation distance and decreasing solution conductivity. There are two effective ways to compensate for this ohmic loss. One is to use a Luggin capillary, which can place the reference electrode sensing point immediately adjacent to the working electrode. Care should be taken to position the capillary tip a little distance from the electrode to avoid shielding interference of the electrode current. The other approach is to use EIS to measure the ohmic loss, and then correct the reported working electrode potential. The multimeter should have a high impedance to prevent data collection from affecting the system.

The energy losses and resistances in BESs are often characterized to better understand the reaction rates occurring at the

electrodes and the mass and charge transfer processes needed to support those reactions. The internal resistance (Ri) can be easily determined from the slope of the linear portion of the polarization curve, ΔE/Δj [43]. This property is critical to system cost, with high Ri systems unable to break even on cost at any current density. It can also be used operationally to optimization power, with the time-dependent Ri used to adjust the external resistance (Rext) for maximum power [44]. Irreversible energy losses associated with different resistances in a BES [45] can be segregated into anode overpotential (ηan), cathode overpotential (ηcat), ionic (Eionic), transport (ET), and pH gradient (EΔpH). The overpotentials can be calculated from Equations 1 and 2 (see attachment), where Ean,measured and Ecat,measured are the measured anode and cathode potentials and Ean and Ecat are the calculated potentials based on the Nernst equation. These calculated potentials are a function of the electrolyte compositions such as buffer type and strength and oxygen concentration. The ionic loss can be calculated based on the electrolyte conductivity. The pH gradient loss across a membrane can be calculated based on the difference between anode and cathode pH. EIS can also be used to estimate resistances based on equivalent circuit analysis. The simplest circuit includes one resistive element, which lumps all the contributions into one parameter. While mass transfer is typically considered to be indicated in a Nyquist plot by a 45º angle in the low-frequency data, it is important to note that mass transfer can be contributing to semicircle features in the plot, particularly in systems with flow. The equivalent circuit model should be appropriate to the system, and the interpretation should be substantiated by other experiments because semicircles alone do not reveal mechanisms.

When conducting BES research, it is important to be mindful of the implications associated with the interdependence between voltage and current and the significance of iR loss. When using a fixed external resistance (Rext), Ean can vary and influence reaction kinetics and possibly microbial competition and community composition. Ean is particularly dynamic in fed-batch systems that are allowed to experience dramatic changes in substrate concentration. Potentiostatic (or galvanostatic) operation remedies this two-variable situation and is recommended for characterizing fundamental BES reactions [46]. In a single-chamber cell with idealized anode and cathode, the relationship between the catalytic current (icat) and the limiting current (iL) is described by Equation 3 (see attachment), where n is the number of electrons involved in the reaction, F is the Faraday's constant, EM is the midpoint potential of the reaction, R is the ideal gas constant, and T is the temperature. In a two-chamber cell, this must be corrected to account for iR loss as shown in Equation 4 (see attachment). From Eq. 4, as iR drop increases the power density curve approaches a parabola, a behavior that is often observed and reported for BES performance. While this describes the system performance, an iR-dominated system masks the electrode behavior. For experiments intended to show differences in electrode performance, this influence can be greatly reduced by using a very small working electrode relative to the counter electrode and membrane. For example, small anodes coupled with large cathodes and membranes were used to compare the performance of different G. sulfurreducens variants [47] and to demonstrate the exoelectrogenic contributions of outer membrane cytochromes and pili [48] in anode-limited conditions.

Cyclic voltammetry (CV) is often used to characterize electrochemically active species, such as mediators or biofilms. Steady-state kinetics should show an overlap in the forward and reverse CV curves. Unlike with electrochemical reactions involving soluble electrode-reactive compounds, reactions in biofilms can take seconds to minutes to achieve steady state due to concentration and potential gradients. Therefore, to characterize steady-state biofilm performance, CV should be performed with a sufficiently slow scan rate as demonstrated by overlapping profiles. Richter et al. [48] observed this behavior with G. sulfurreducens biofilms at 0.002 V/s. The hysteresis observed at higher CV scan rates can reveal additional information about the mechanisms and relative rates of electron transfer from the non-electrode-reactive substrate through the biofilm to the electrode [48, 49].

There are several methods reported in the BES literature to generate polarization data, from which power density curves are constructed. The most common approach has been the stepwise change of Rext and the collection of the stable voltage at each Rext. In fed-batch systems, this can be done within a single cycle, typically waiting at least 20 minutes at each Rext. This method requires sufficient initial substrate concentration and buffer intensity to avoid substrate- or pH-limited conditions in the latter Rext readings. Batch profiles often do not show steady performance, which becomes problematic with respect to when to collect measurements during the cycle. Also, "power overshoot" is often evident in data collected with this method, when the cell voltage drops rapidly at lower Rext and sometimes even supports a lower current density [50]. This signature power overshoot is called Type D, and the cell voltage change is caused by a rapid increase in Ean. It can be solved by allowing the biofilm longer acclimation times to the low Rext¬ conditions, such as running multiple cycles at each condition [51]. Even when Type D power overshoot is evident, all of the polarization data should be reported including the points at which the power doubles back. Another form of power overshoot, called Type M, occurs when the maximum power density is exaggerated due to an overestimation of Ecat. This is observed when the system is not given sufficient time to stabilize, such as when linear sweep voltammetry (LSV) is used at too high a rate to collect polarization data. Type M overshoot can be remedied by slowing down the scan rate until there is no scan-rate dependency of the polarization response, or simply by performing the single- or multiple-cycle varied resistance method [52]. The common experience of the workshop participants to eliminate Type M overshoot was to use an LSV scan rate of 0.1 mV/s, but this should be demonstrated for the specific system of interest.

Standard Reporting of Performance Metrics for Comparative Evaluations

Power density is one of the main performance metrics used to compare different BESs. Measurements should be conducted to avoid power overshoot artifacts, as mentioned above. In addition, there are a number of ways to normalize the power from a system to allow comparisons among different design platforms. It is best to report power density with respect to

both rate-limiting electrode area and empty-bed reactor volume, or equivalently to provide the area to volume ratio, since this ratio does not necessary remain constant during system modifications or scale up. For example, in the standard 28-ml air-cathode cubic MFC with 4 cm separation between electrodes [53], the electrode area to volume ratio is 25 m2/m3. The 2 -cm design doubles this ratio to 50 m2/m3. A mini-reactor design that allows inexpensive parallel testing of many reactors has ratios of 92 m2 anode/m3 and 86 m2 cathode/m3 [54]. Clearly the details on reactor geometry must be fully reported to allow equivalent comparisons. Another suggestion was to report the energy produced or consumed normalized by the substrate concentration (e.g. kJ/g COD (chemical oxygen demand) or kWh/g COD) to capture system performance within the context of the substrate addition.

Many BES studies are conducted with fed-batch systems, which often show irregular voltage or current profiles during a cycle that lack a clear performance plateau or stable condition. This non-steady-state batch condition complicates the meaningful characterization of performance, as the peak voltage or current in the cycle may be very transient and uncharacteristic of the average. One approach for handling this situation and allowing the comparison of very dissimilar profiles is to report the total coulomb recovery. Coulombic efficiency (CE) can be calculated based on the input COD, which does not account for the residual substrate that was not degraded, or the change in COD during a cycle, which captures the fraction of electrons that the microbes extracted from the substrate. The suggestion was to refer to these two metrics differently, using the term CE when dividing by the change in COD and the term Coulombic recovery when using the total input COD.

As mentioned above, solution chemistry can have a strong impact on system performance. One parameter that has a marked impact on performance and must always be reported is solution conductivity [55]. This is not only critical for the comparison of results from different studies, but also to allow proper comparison of different treatments within a single study. It is not uncommon for papers to compare the performances of different treatments, for example diluted wastewater or different pH, without reporting the conductivity effects of the treatments. A preferable approach is to adjust all treatments to a common conductivity, to eliminate that effect [56]. Also, while acetate makes a great simple substrate for testing and perhaps even benchmarking the performance of different reactor designs, it should never be discussed as a wastewater surrogate as the performance is marked different [57]. Oxygen intrusion is another solution chemistry issue that can significantly affect performance. For example, microaerophilic conditions have been shown to cause increased current production in Pseudomonas aeruginosa PA14 through a quorum sensing regulatory cascade [58]. However, this issue is rarely accounted for or acknowledged. Strictly anaerobic conditions can be maintained by using an overpressure of nitrogen.

Replication of BES performance is another issue that is inconsistent and often lacking in the literature. This can be achieved by running replicate parallel systems (biological replicates) and replicate monitoring of a single system (technical replicates). Continuous-flow reactors can sometimes show better reproducibility than replicate fed-batch systems.

Alternative Applications

There are numerous BES applications being developed that incorporate some additional functionality into a BES. The performance metrics of this addition feature will be dependent on the application. For example, hydrogen production in a microbial electrolysis cell (MEC) would be characterized by a specific production rate, such as m3 H2/m3/d. In microbial desalination cells (MDCs), which couple the function of desalination to the microbe-induced current of a BES, it is insufficient to just report the percent total dissolved solids (TDS) removal. The removal needs to be expressed as a rate (g TDS/L/d) to allow a more meaningful performance comparison. In all BES variants, the unifying feature is current density, so this is proposed as the metric that crosses application and design platforms.

It becomes more difficult to identify meaningful and equivalent metrics that allow comparison of BESs with their more traditional technology counterpart. For example, BESs designed for wastewater treatment should be compared to aerobic processes such as activated sludge, with the need to achieve the treatment performance constraints. BESs for energy production could be compared with anaerobic digestion. MDCs should be compared with reverse osmosis systems. Bioremediation applications of BESs could be compared with direct electron donor or electron acceptor additions. One possible tool for comparing BESs with alternate technologies is the use of life cycle analysis.

For BES applications that generate a product (e.g., hydrogen, 1,3-propanediol, caustic soda [59], or hydrogen peroxide [60]), the assessment of economic feasibility requires a comparison of inputs (e.g., substrate, energy if necessary, and capital costs) and outputs (e.g., product quantity and quality). If the product value is less than the input costs, BESs are not suitable for this application. In terms of substrate input, the energy of wastewater is very difficult to determine, and in the context of product generation the treatment efficiency is probably not the focus of performance. In this case, CE is the relevant measure of input substrate utilization. For capital costs, a membrane electrode assembly (MEA) configuration has equivalent projected surface areas for the anode, membrane, and cathode, and they are sold per area. Hence, the primary electrochemical performance metric is the current density expressed per MEA area. For product output, one could calculate the energy content of the product (kJ/mol), but more functionally useful metrics would be the CE of product recovery and the specific energy input (kJ/kg product). The quality and quantity of the product are also critical parameters, as they affect downstream processing and utilization. Quality should be expressed in terms of product concentration (or titer) and purity. Quantity is the production rate in g product/L/hr. The electron efficiency, which is the electrons from the substrate that are recovered in the product, is a particularly useful metric in that it allows an easy comparison of different substrates, products, and costs.

Workshop Outcomes

The primary outcomes of the workshop were the cumulative recommendations for benchmarking, monitoring, and reporting

BES system performance that were developed during the open discussions (Appendix 2). Given the diversity of BES applications and various foci in BES studies, and in some cases an inability to come to full agreement due to the need for continued research, it was not possible to develop a fixed set of conditions that could be applied to all BES studies and demonstrations. Rather, a list was developed with appropriate qualifiers to acknowledge and account for these different considerations.

A second indirect outcome of the workshop, derived from bringing many of the leading BES researchers together, was the development of a professional society called the International Society of Microbial Electrochemical Technologies (ISMET). Since the workshop, the board of directors has developed and approved bylaws, started the development of a website, and begun to advertise this professional organization for those interested in BESs. The society has sections for North America (NA-ISMET), Europe (EU-ISMET), and Asia-Pacific (AP-ISMET).

Acknowledgments

Funding for the workshop was provided by the Army Research Office and the U.S. Army Engineer Research and Development Center under Award W911NF-11-10148.

List of Attendees

Mr. Jack Ambler, Siemens; Dr. Lars Angenent, Cornell University; Dr. Haluk Beyenal, Washington State University; Dr. Wally Buchholz, Life Sciences Division, U.S. Army Research Office; Dr. Doug Call, Penn State University; Mr. John Cirucci, Air Products and Chemicals Inc.; Mr. Roland Cusick, Penn State University; Ms. Caroline Dale, Veolia Water Systems in USA; Dr. Pat Evans, CDM; Dr. Ashley Franks, University of Massachusetts / Latrobe University – Melbourne; Dr. Bert Hamelers, Wageningen University; Dr. Zhen He, University of Wisconsin – Milwaukee; Dr. Mike Hickner, Penn State University; Dr. Karl Indest, Environmental Microbiology Team Leader, US Army Corps of Engineers; Dr. Pat Kiely, Cambrian Innovation; Dr. Hong Liu, Oregon State University; Dr. Bruce Logan, Penn State University; Mr. Jerry Miller, ERDC's Assistant to the Technical Director for Military Environmental Engineering and Sciences; Dr. Kurt Preston, Chief, Environmental Sciences Division, Army Research Office; Dr. Korneel Rabaey, University of Queensland / Ghent University; Dr. Jay Regan, Penn State University; Dr. Zhiyong Ren, University of Colorado – Denver; Mr. Ryan Restow, Washington State University; Dr. Tom Richard, Penn State University; Mr. Dave Ringelberg, Microbiologist, US Army ERDC-CRREL; Dr. Jim Sumner, Research Chemist / Team Leader, US Army Research Laboratory; Dr. Boris Tartakovsky, Canadian Research Council; Dr. Lenny Tender, Center for Bio/Molecular Science and Engineering, US Naval Research Laboratory; Dr. César Torres, Arizona State University; Ms. Rachel Wagner, Penn State University.

References

1. Logan, B.E., Exoelectrogenic bacteria that power microbial fuel cells. Nature Reviews Microbiology, 2009. 7(5): p. 375-381.
2. Min, B., S. Cheng, and B.E. Logan, Electricity generation using membrane and salt bridge microbial fuel cells. Water Research, 2005. 39(9): p. 1675-1686.
3. Logan, B.E., Scaling Up Microbial Fuel Cells and Other Bioelectrochemical Systems. Applied Microbiology and Biotechnology, 2010. 85(6): p. 1665-1671.
4. Kiely, P.D., et al., Long-term cathode performance and the microbial communities that develop in microbial fuel cells fed different fermentation endproducts. Bioresource Technology, 2011. 102(1): p. 361-366.
5. Larrosa-Guerrero, A., et al., Open circuit versus closed circuit enrichment of anodic biofilms in MFC: effect on performance and anodic communities. Applied Microbiology and Biotechnology, 2010. 87(5): p. 1699-1713.
6. Jung, S. and J.M. Regan, Influence of External Resistance on Electrogenesis, Methanogenesis, and Anode Prokaryotic Communities in Microbial Fuel Cells. Applied and Environmental Microbiology, 2011. 77(2): p. 564-571.
7. Ren, Z.Y., et al., Characterization of Microbial Fuel Cells at Microbially and Electrochemically Meaningful Time scales. Environmental Science & Technology, 2011. 45(6): p. 2435-2441.
8. Kiely, P.D., et al., Anode microbial communities produced by changing from microbial fuel cell to microbial electrolysis cell operation using two different wastewaters. Bioresource Technology, 2011. 102(1): p. 388-394.
9. Crosby, L.D. and C.S. Criddle, Understanding bias in microbial community analysis techniques due to rrn operon copy number heterogeneity. Biotechniques, 2003. 34(4): p. 790-+.
10. Jung, S.H. and J.M. Regan, Comparison of Anode Bacterial Communities and Performance in Microbial Fuel Cells with Different Electron Donors. Applied Microbiology and Biotechnology, 2007. 77(2): p. 393-402.
11. Ren, Z.Y., et al., Time-course correlation of biofilm properties and electrochemical performance in single-chamber microbial fuel cells. Bioresource Technology, 2011. 102(1): p. 416-421.
12. Xing, D.F., et al., Change in microbial communities in acetate- and glucose-fed microbial fuel cells in the presence of light. Biosensors & Bioelectronics, 2009. 25(1): p. 105-111.
13. Hughes, J.B. and J.J. Hellmann, The application of rarefaction techniques to molecular inventories of microbial diversity. Environmental Microbiology, 2005. 397: p. 292-+.
14. Kemp, P.F. and J.Y. Aller, Estimating prokaryotic diversity: When are 16S rDNA libraries large enough? Limnology and Oceanography-Methods, 2004. 2: p. 114-125.
15. Lee, T.K., et al., Discovery of commonly existing anode biofilm microbes in two different wastewater treatment MFCs using

FLX Titanium pyrosequencing. Applied Microbiology and Biotechnology, 2010. 87(6): p. 2335-2343.
16. Lu, L., D. Xing, and N. Ren, Pyrosequencing reveals highly diverse microbial communities in microbial electrolysis cells involved in enhanced H2 production from waste activated sludge. Water Research, 2012. 46: p. 2425-2434.
17. Zhang, G.D., et al., Biocathode microbial fuel cell for efficient electricity recovery from dairy manure. Biosensors & Bioelectronics, 2012. 31(1): p. 537-543.
18. Ren, Z., L.M. Steinberg, and J.M. Regan, Electricity Production and Microbial Biofilm Characterization in Cellulose-Fed Microbial Fuel Cells. Water Science and Technology, 2008. 58(3): p. 617-622.
19. He, Z., S.D. Minteer, and L.T. Angenent, Electricity generation from artificial wastewater using an upflow microbial fuel cell. Environmental Science & Technology, 2005. 39(14): p. 5262-5267.
20. Chung, K. and S. Okabe, Continuous power generation and microbial community structure of the anode biofilms in a three-stage microbial fuel cell system. Applied Microbiology and Biotechnology, 2009. 83(5): p. 965-977.
21. Xing, D., et al., Isolation of the exoelectrogenic denitrifying bacterium Comamonas denitrificans based on dilution-to-extinction of the microbial community. Applied Microbiology and Biotechnology, 2010. 85(5): p. 1575-1587.
22. Yates, M.D., et al., Convergent development of anodic bacterial communities in microbial fuel cells. The ISME Journal, 2012. In print.
23. Vargas, I.T., I.U. Albert, and J.M. Regan, Physical Distribution of Bacterial Communities on Volumetric and Planar Anodes in Single-Chamber Air-Cathode Microbial Fuel Cells. In preparation, 2012.
24. Franks, A.E., N. Malvankar, and K.P. Nevin, Bacterial biofilms: the powerhouse of a microbial fuel cell. Biofuels, 2010. 1(4): p. 589-604.
25. Reguera, G., et al., Biofilm and nanowire production leads to increased current in Geobacter sulfurreducens fuel cells. Applied and Environmental Microbiology, 2006. 72: p. 7345-7348.
26. Ren, Z., et al., Characterization of the Cellulolytic and Hydrogen-Producing Activities of Six Mesophilic Clostridium Species. Journal of Applied Microbiology, 2007. 103(6): p. 2258-2266.
27. Nevin, K.P., et al., Anode Biofilm Transcriptomics Reveals Outer Surface Components Essential for High Density Current Production in Geobacter sulfurreducens Fuel Cells. Plos One, 2009. 4(5).
28. Nevin, K.P., et al., Microbial Electrosynthesis: Feeding Microbes Electricity To Convert Carbon Dioxide and Water to Multicarbon Extracellular Organic Compounds. Mbio, 2010. 1(2).
29. Strycharz, S.M., et al., Reductive dechlorination of 2-chlorophenol by Anaeromyxobacter dehalogenans with an electrode serving as the electron donor. Environmental Microbiology Reports, 2010. 2(2): p. 289-294.
30. Strycharz, S.M., et al., Graphite electrode as a sole electron donor for reductive dechlorination of tetrachlorethene by Geobacter lovleyi. Applied and Environmental Microbiology, 2008. 74(19): p. 5943-5947.
31. Nevin, K.P., et al., Electrosynthesis of Organic Compounds from Carbon Dioxide Is Catalyzed by a Diversity of Acetogenic Microorganisms. Applied and Environmental Microbiology, 2011. 77(9): p. 2882-2886.
32. Franks, A.E., et al., Microtoming coupled to microarray analysis to evaluate the spatial metabolic status of Geobacter sulfurreducens biofilms. ISME Journal, 2010. 4(4): p. 509-519.
33. Franks, A.E., et al., Novel strategy for three-dimensional real-time imaging of microbial fuel cell communities: monitoring the inhibitory effects of proton accumulation within the anode biofilm. Energy and Environmental Science, 2009. 2: p. 113–119.
34. Malvankar, N.S., et al., Tunable metallic-like conductivity in microbial nanowire networks. Nature Nanotechnology, 2011. 6(9): p. 573-579.
35. Butler, C.S. and R. Nerenberg, Performance and microbial ecology of air-cathode microbial fuel cells with layered electrode assemblies. Applied Microbiology and Biotechnology, 2010. 86(5): p. 1399-1408.
36. Babauta, J.T., H.D. Nguyen, and H. Beyenal, Redox and pH Microenvironments within Shewanella oneidensis MR-1 Biofilms Reveal an Electron Transfer Mechanism. Environmental Science & Technology, 2011. 45(15): p. 6654-6660.
37. Nguyen, H.D., et al., A voltammetric flavin microelectrode for use in biofilms. Sensors and Actuators B: Chemical, 2012. 161(1): p. 929–937.
38. Renslow, R.S., et al., In Situ Effective Diffusion Coefficient Profiles in Live Biofilms Using Pulsed-Field Gradient Nuclear Magnetic Resonance. Biotechnology and Bioengineering, 2010. 106(6): p. 928-937.
39. Marsili, E., et al., Microbial Biofilm Voltammetry: Direct Electrochemical Characterization of Catalytic Electrode-Attached Biofilms. Applied and Environmental Microbiology, 2008. 74(23): p. 7329-7337.
40. Jung, S., M.M. Mench, and J.M. Regan, Impedance Characteristics and Polarization Behavior of a Microbial Fuel Cell in Response to Short-Term Changes in Medium pH. Environmental Science & Technology, 2011. 45(20): p. 9069-9074.
41. Saito, T., et al., Investigation of ionic polymer cathode binders for microbial fuel cells. Electrochimica Acta, 2010. 55(9): p. 3398-3403.
42. Saito, T., et al., Neutral hydrophilic cathode catalyst binders for microbial fuel cells. Energy & Environmental Science, 2011. 4(3): p. 928-934.
43. Ter Heijne, A., et al., Copper Recovery Combined with Electricity Production in a Microbial Fuel Cell. Environmental Science & Technology, 2010. 44(11): p. 4376-4381.
44. Woodward, L., et al., Comparison of real-time methods for maximizing power output in microbial fuel cells. AIChE Journal, 2010. 56(10): p. 2742-2750.
45. Sleutels, T.H.J.A., et al., Ion transport resistance in Microbial Electrolysis Cells with anion and cation exchange membranes. International Journal of Hydrogen Energy, 2009. 34(9): p. 3612-3620.

46. Z, L., et al., Bacteria-based AND logic gate: a decision-making and self-powered biosensor. Chemical Communications, 2011. 47: p. 3060-3062.
47. Yi, H.N., et al., Selection of a variant of Geobacter sulfurreducens with enhanced capacity for current production in microbial fuel cells. Biosensors & Bioelectronics, 2009. 24(12): p. 3498-3503.
48. Richter, H., et al., Cyclic voltammetry of biofilms of wild type and mutant Geobacter sulfurreducens on fuel cell anodes indicates possible roles of OmcB, OmcZ, type IV pili, and protons in extracellular electron transfer. Energy & Environmental Science, 2009. 2(5): p. 506-516.
49. Strycharz, S.M., et al., Application of cyclic voltammetry to investigate enhanced catalytic current generation by biofilm-modified anodes of Geobacter sulfurreducens strain DL1 vs. variant strain KN400. Energy & Environmental Science, 2011. 4(3): p. 896-913.
50. Watson, V.J. and B.E. Logan, Analysis of polarization methods for elimination of power overshoot in microbial fuel cells. Electrochemistry Communications, 2011. 13(1): p. 54-56.
51. Hong, Y.Y., et al., Adaptation to high current using low external resistances eliminates power overshoot in microbial fuel cells. Biosensors & Bioelectronics, 2011. 28(1): p. 71-76.
52. Velásquez-Orta, S.B., T.P. Curtis, and B.E. Logan, Energy from algae using microbial fuel cells. Biotechnology and Bioengineering, 2009. 103(6): p. 1068-1076.
53. Liu, H. and B.E. Logan, Electricity Generation Using an Air-Cathode Single Chamber Microbial Fuel Cell in the Presence and Absence of a Proton Exchange Membrane. Environmental Science & Technology, 2004. 38(14): p. 4040-4046.
54. Call, D.F. and B.E. Logan, A method for high throughput bioelectrochemical research based on small scale microbial electrolysis cells. Biosensors & Bioelectronics, 2011. 26(11): p. 4526-4531.
55. Nam, J.-Y., et al., Variation of power generation at different buffer types and conductivities in single chamber microbial fuel cells. Biosensors & Bioelectronics, 2010. 25(5): p. 1155–1159.
56. Feng, Y., X. Wang, and B.E.L.H. Lee, Brewery wastewater treatment using air-cathode microbial fuel cells. Applied Microbiology amd Biotechnology, 2008. 78(5): p. 873-880.
57. Hays, S., F. Zhang, and B.E. Logan, Performance of two different types of anodes in membrane electrode assembly microbial fuel cells for power generation from domestic wastewater. Journal of Power Sources, 2011. 196(20): p. 8293- 8300.
58. Venkataraman, A., et al., Quorum sensing regulates electric current generation of Pseudomonas aeruginosa PA14 in bioelectrochemical systems. Electrochemistry Communications, 2010. 12(3): p. 459-462.
59. Rabaey, K., et al., High Current Generation Coupled to Caustic Production Using a Lamellar Bioelectrochemical System. Environmental Science and Technology, 2010. 44(11): p. 4315-4321.
60. Rozendal, R.A., et al., Efficient hydrogen peroxide generation from organic matter in a bioelectrochemical system. Electrochemistry Communications, 2009. 11: p. 1752–1755

Technology Transfer

Workshop Report - Attachment

Bioelectrochemical Systems Workshop:
Standardized Analyses, Design Benchmarks, and Reporting

John M. Regan
Department of Civil and Environmental Engineering, The Pennsylvania State University, University Park, Pennsylvania, USA

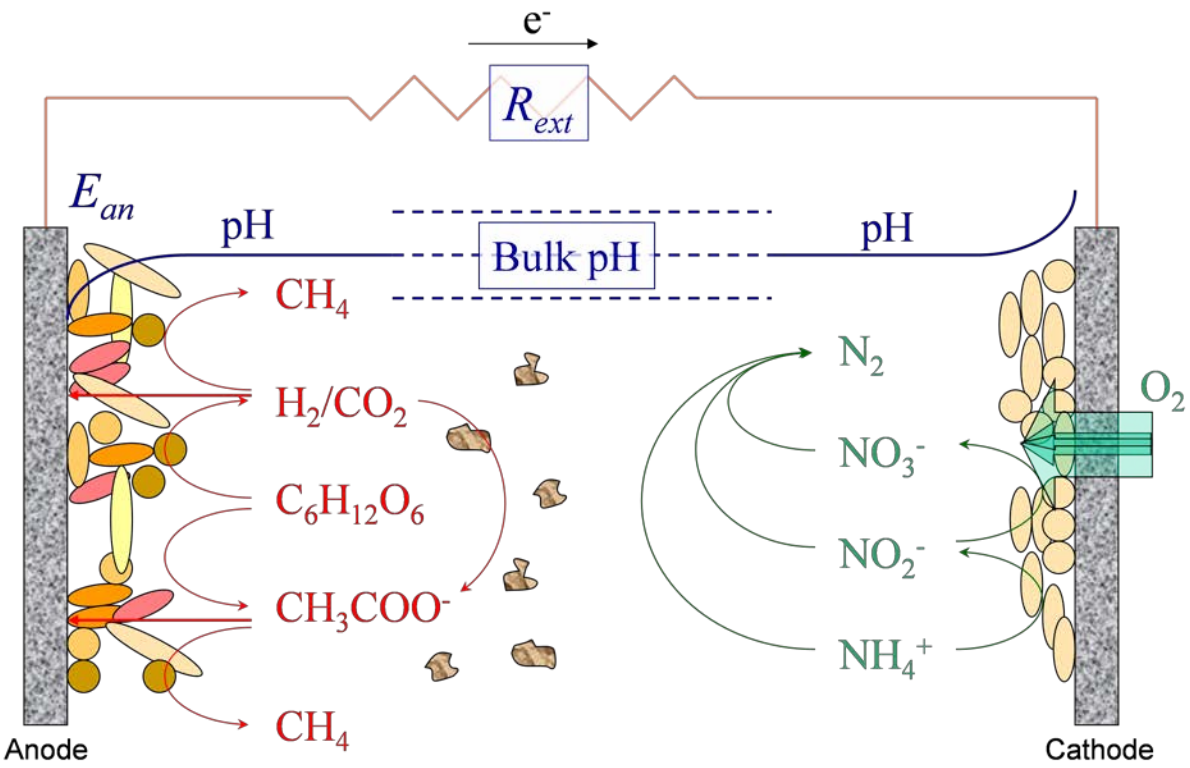

Figure 1. Ecology in mixed-culture air-cathode microbial fuel cell and several influencing factors.

Equations:

$$\eta_{an} = E_{an,measured} - E_{an} \quad (1)$$

$$\eta_{cat} = E_{cat,measured} - E_{cat} \quad (2)$$

$$i_{cat} = i_L \left[\frac{\left(e^{\frac{nF(E-E_M)}{RT}} \right)}{\left(1+e^{\frac{nF(E-E_M)}{RT}} \right)} \right] \quad (3)$$

$$i_{cat} = i_L \left[\frac{\left(e^{\frac{nF((E-iR)-E_M)}{RT}} \right)}{\left(1+e^{\frac{nF((E-iR)-E_M)}{RT}} \right)} \right] \quad (4)$$

Appendix 1. Workshop agenda

Wednesday, September 14th

8:45 AM Jerry Miller, ERDC's Assistant to the Technical Director for Military Environmental Engineering and Sciences, US Army Corps of Engineers,

9:00 AM Jay Regan, Penn State University
Microbial community characterizations

9:20 AM Ashley Franks, University of Massachusetts / Latrobe University - Melbourne
Microbial considerations

9:40 AM Haluk Beyenal, Washington State University
Biofilm characterization using microelectrodes and microscale processes in biofilms

10:00 AM Mike Hickner, Penn State University
Characterizing electrode binders and polymers for MFCs

10:20 AM Break

10:40 AM César Torres, Arizona State University
Anode kinetics and potentials

11:00 AM Bert Hamelers, Wageningen University
Mass/charge transfer

11:20 AM Lenny Tender, US Naval Research Laboratory
Analytical electrochemistry

11:40 AM Hong Liu, Oregon State University
Evaluation of MFC performance using linear sweep voltammetry

12:00 PM Lunch

1:00 PM Zhen He, University of Wisconsin – Milwaukee
Determining power and efficiency

1:20 PM Boris Tartakovsky, Canadian Research Council
Real-time control and modeling techniques in standardizing/ benchmarking BESs

1:40 PM Korneel Rabaey, University of Queensland / Ghent University
Expressing product formation in relevant terms for the (bio)production industry

2:00 PM Zhiyong Ren, University of Colorado – Denver
Different functions in BESs

2:20 PM Bruce Logan, Penn State University
Reactor architecture and stacks

2:40 PM Largus Angenent, Cornell University
Replicates, O_2 influence, continuous vs batch

3:00 PM Jim Sumner, US Army Research Laboratory
Standardization in the ARL, pros and cons

3:20 PM Break

3:30 PM Group discussions

Topics: (1) Benchmark designs
(2) Monitoring/characterization techniques
- microbiology
- materials
- electrochemistry
- solution/gas chemistry

5:00 PM Adjourn

Thursday, September 15th

8:30 AM Continue group discussions

Topics: (3) Reporting requirements
(4) Unique considerations for non-MFC systems
(5) What are we not ready to standardize?

Appendix 2. Recommendations for benchmarking, monitoring, and reporting[1]

Benchmarks:

- Nafion binder
- Temperature (not specified)
- Electrodes
    - Glassy carbon
    - 0.5 mg/cm^2 Pt with Nafion cathode
- Microbial benchmark
    - *Geobacter* - caution of reactor effects and O_2 intrusion
    - *Shewanella* (O_2 tolerance, mediator)
- Medium
    - acetate
    - conductivity
- Reactor (single chamber)
- 2-electrode reactors
- Single 3-electrode cells
- Performance compared to conventional technology (e.g., anaerobic digestion for microbial fuel cell or microbial electrolysis cell, and reverse osmosis for microbial desalination cell), KW/KJ consumed

Monitoring / characterization /operational techniques:

Biological

- Community analysis – caution with quantitative comments, statistical analysis needed, appropriate electrode for the objectives
- Pure culture confirmation – PCR, plate
- Frozen stocks for each test (pure and mixed cultures, spin/wash glycerol/resuspend)
- Concentration and activity (ATP, BactiQuant, BacLight [caution on applicability], protein, DNA)

Materials

- Surface area (ferricyanide, BET, Ar – non exactly capture microbially accessible electrochemically active surface area)
- Membranes (IEC, water content) <- requires materials science input

Feedstocks – no consensus here, too application dependent

Electrochemistry

- Reference electrode – Ag/AgCl conversion, calculate drift for your medium, caution about variable distance from working electrode and non-standard geometry, procedure to correct for ohmic resistance, correction for temperature
- Cyclic voltammetry (CV) – use for demonstrating chemical steady state
- Linear sweep voltammetry (LSV) – scan rate (slow, confirm response is independent of scan rate if chemical steady state is the goal)
- Chronoamperometry

- Polarization – show all the data and approaching i_{max}, high-to-low R, overcome overshoot by acclimating at low R_{ext}, include E_{an} and E_{cat}, time considerations (10-30 min, minimal drift), low R requires good resistor box and meter
- For batch systems, need confirmation of minimal change in S
- R_{int} – electrochemical impedance spectroscopy (EIS) at polarization curve midpotential, linear portion of polarization curve (define!). Current interrupt not recommended based on insufficient validation
- EIS – Nyquist, Bode, show model fit
- Statistics/replication

Solution/gas chemistry

- Coulombic efficiency (CE) – Respirometric CE (current/consumed COD), Capture CE (current/total COD), Efficiency is (in – out), Recovery is in
- Energy efficiency, energy recovery, specific energy (J/g), specific rate (J/g/d), specific energy loading rate (e.g., g $COD_{in}/m^2/d$)
- Account for energy quality
- Concentration/purity of product (e.g., H_2, CH_4, other product, desalinated water) should be characterized

Reporting:

Chemistry/conditions

- Alkalinity
- Salts (Ca, Mg,)
- Trace metals
- Medium composition
- Sterility
- Buffer composition/capacity
- Conductivity
- TOC, DOC
- COD (influent, effluent) - total, soluble
- BOD
- pH (initial and final)
- Temperature
- Rf
- DO
- TDS
- CH_4, H_2, CO_2 (concentration, production rate)
- Redox solution
- E_{an}/E_{cat}

Operation

- Continuous versus batch
- Retention time
- Re
- Flow rate
- Anaerobic conditions… feed prep, degassed, etc.

- Duration
- Demonstration of steady state

Design
- Area (electrodes, membrane)
- Volume (empty bed)
- Electrode spacing (other dimensions, shape?)
- Electrode preparation
- Materials (electrode/membrane)
- Number of chambers

Performance
- I
- R_{int}
- CE

Feedstock
- Wastewater characterizations (VFAs, TKN, NO_3^-, SO_4^-, …)

Biomass
- Concentration (e.g., protein)
- Inoculum (acclimated or unacclimated, antecedent conditions, electrode potential)
- Strategy (multiple, volume %)

Community composition – no consensus on method, but multiple methods preferred

[1] The workshop did not have electrical engineers and material scientists represented, so power conditioning and materials considerations are not adequately addressed in these recommendations.